

CLAIMS

1. A method for the detection of a pathogenic form of a prion protein in a sample, comprising providing a container, pretreating the container to deposit on a surface of the container a coating of a cellulose derivative capable of favoring the binding of pathogenic prion protein to said container surface over
5 the binding of cellular prion protein, incubating the sample in said container to bind any pathogenic prion protein present in the sample to said container surface, labelling the thus immobilized pathogenic prion protein, if present, with an appropriate labelling agent using an anti-prion antibody capable of binding to pathogenic prion protein and detecting the presence of labelling agent attached
10 to the container surface.
2. A method according to claim 1, which is an enzyme-linked immunosorbent assay (ELISA) using a labelling enzyme as labelling agent, wherein the presence of labelling enzyme attached to the container surface is detected by incubating the container with an appropriate substrate for the labelling enzyme and
15 detecting conversion of the substrate into a coloured, fluorescent or luminescent product.
3. A method according to claim 1 or 2, wherein the cellulose derivative used favors the binding of pathogenic prion protein over cellular prion protein by enhancing the binding of pathogenic prion protein to the container surface, or
20 reducing the binding of cellular prion protein to the container surface, or both.
4. A method according to any one of claims 1-3, wherein the container is transparent.
5. A method according to any one of claims 1-4, wherein the container is a well of a microtitre plate.
- 25 6. A method according to any one of claims 1-5, wherein the coating of a cellulose derivative deposited on a surface of the container is transparent.
7. A method according to any one of claims 1-6, wherein the cellulose derivative used is insoluble in water.
8. A method according to claim 7, wherein the cellulose derivative is a
30 nitrocellulose.
9. A method according to any one of claims 1-8, wherein said pretreatment comprises incubating the container with a solution of an appropriate cellulose

derivative in a non-aggressive solvent, such as methanol or ethanol, followed by evaporation of the solvent.

10. A method according to claim 9, wherein the solution contains from 0.001 to 20 mg/ml of the cellulose derivative.

5 11. A method according to any one of claims 1-10, wherein the cellulose derivative is deposited onto the surface of the container in an amount of from 1 to 20,000 ng/mm².

12. A method according to any one of claims 1-11, wherein the sample is pretreated with an enzyme capable of digesting cellular prion protein.

10 13. A method according to claim 12, wherein the enzyme is proteinase K.

14. A method according to claim 12 or 13, wherein the enzymatic digestion is stopped by briefly heating the sample to a temperature of from 70 to 100 °C to inactivate the enzyme.

15 15. A method according to claim 14, wherein the sample after the inactivation of the enzyme is treated with detergent at a temperature of from 70 to 100 °C to denature the protein in the sample.

16. A method according to any one of claims 12-15, wherein the pretreatment of the container with a cellulose derivative favors the binding of enzymatically pretreated pathogenic prion protein over enzymatically pretreated cellular prion protein by enhancing the binding of enzymatically pretreated pathogenic prion protein to the container surface, or reducing the binding of enzymatically pretreated cellular prion protein to the container surface, or both.

20 17. A method according to any one of claims 1-16, wherein the labelling of any pathogenic prion protein immobilized onto the container surface is performed either directly with an enzyme-labelled anti-prion antibody, or indirectly with a non-labelled anti-prion antibody followed by an enzyme-labelled antibody capable of binding to the anti-prion antibody.

18. A method according to any one of claims 1-17, wherein a peroxidase, such as Horse Radish Peroxidase, is used as a labelling enzyme.

30 19. A method according to any one of claims 1-18, wherein 3,3',5,5'-tetramethylbenzidine is used as a substrate for the labelling enzyme.

20. A method according to any one of claims 1-19, wherein the substrate conversion results in a coloured material which is detected by direct readout of the absorbance.

35 21. A method according to any one of claims 1-20, wherein the quantitative occurrence of a pathogenic form of a prion protein in a sample is determined.

22. A method according to any one of claims 1-21, wherein the ELISA includes a parallel ELISA without pretreatment of the container as a control of complete enzymatic digestion of cellular prion protein.
23. A method according to any one of claims 1-22, wherein the sample is
5 derived from brain or lymphoid tissue of a human or animal.
24. A method according to any one of claims 1-22, wherein the sample is a body fluid, such as blood, plasma, cerebrospinal fluid, saliva, sputum, seminal fluid, vaginal fluid or urine.
25. A method for separating pathogenic prion protein from a mixture which
10 contains pathogenic prion protein and cellular prion protein comprising contacting the mixture with a surface which preferentially binds pathogenic prion protein and separating the non-bound material from said surface.
26. A method according to claim 25, wherein said surface is made of, or coated with, a water-insoluble cellulose derivative, such as nitrocellulose.
- 15 27. A method according to claim 25 or 26, wherein said surface is a surface of a microtitre plate well coated with a layer of nitrocellulose.
28. A method according to claim 25 or 26, wherein said surface is a surface of a bead or column filling which is made of, or coated with, nitrocellulose.
29. A method according to any one of claims 25-28, wherein said mixture is
20 pretreated with an enzyme capable of digesting cellular prion protein, such as proteinase K.
30. A method according to claim 29, wherein after the enzymatic digestion first the digestion enzyme is inactivated and then the protein which is present in the mixture is denatured.